# Using Ismeans

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#### Abstract

Least-squares means are predictions from a linear model, or averages thereof. They are useful in the analysis of experimental data for summarizing the effects of factors, and for testing contrasts among certain marginal predictions. The **lsmeans** package provides a simple and rather comprehensive formula-based way of specifying least-squares means and contrasts thereof. It supports most R packages that fit linear or mixed models.

Keywords: least-squares means, linear models, experimental design.

## 1. Introduction

### 1.1. What are least-squares means?

Least-squares means (or LS means), are generalizations of covariate-adjusted means, and date back at least to 1976 when they were incorporated in the contributed SAS procedure named HARVEY (Harvey 1976). Later, they were incorporated via LSMEANS statements in the regular SAS releases. SAS's documentation describes them as "predicted population margins—that is, they estimate the marginal means over a balanced population" (SAS Institute Inc. 2012).

People disagree on the appropriateness of LS means. As in many statistical calculations, there are times when they are appropriate, and times when they are not. However, as long as one understands what is being calculated, one can judge its appropriateness. The main thing to remember is that LS means are simply predictions from a model over a grid of predictor values, or marginal averages thereof. More explicitly, define a set of reference levels for each predictor, and create a grid (call it the reference grid) consisting of all combinations of these. Make predictions on this grid, and compute marginal means of those predictions, if needed (usually using equal weights). For clarity, we refer to these averaged predictions as marginal LS means.

The default in **Ismeans** is to set the reference levels as follows: For predictors of class **factor** or **ordered**, the default reference levels are the levels of the factor. For numeric predictors, the default is to use a single reference level at the mean value of the predictor. It is possible to change the reference levels, and if this is done, it is extremely important to understand that this also alters the definition of any marginal LS means, as the averaging is done over a different set of levels.

### 1.2. Package overview

The Ismeans package (Lenth 2014) is built upon objects of class ref.grid which defines the grid of reference levels to use for the predictions. Such ref.grid objects are provided for linear models produced by most linear-models functions including 1m and aov in the stats package; 1me and gls from the nlme package (Pinheiro, Bates, and R-core 2013); and 1mer and others from the lme4 package (Bates, Maechler, Bolker, and Walker 2013). aov is supported only if the model does not contain an Error() term. Generalized linear models and GLMMs are also supported, where LS means are defined in terms of the linear predictor (before applying the link function). For 1m objects, special provisions are included to check for estimability when the model is rank-deficient. Provisions are also made for models with a multivariate response, so that the dimensions of the response can be specified in the same way as the levels of a factor.

As explained before, LS means are predictions over the reference grid, or marginal averages thereof. These are computed by the function lsmeans, which works with either a ref.grid or a model object. The desired sets of LS means are specified using the names of the predictors, and optionally the names of "by" variables for grouping. Alternatively, these can be specified using a formula, e.g., ~ dose | treat requests the LS means for each dose, within each treatment. lsmeans creates an object of class lsmobj, a sub-class of ref.grid.

The summary method for ref.grid and lsmobj objects computes estimates, standard errors, confidence intervals, test statistics, and P values. It also allows for groupings by one or more variables, and allows for various adjustments for multiplicity of tests.

There are several useful functions that can be used to do follow-up analyses. The most important one is contrast, which computes contrasts of LS means. A number of standard contrast families are provided and they can be specified by name, e.g., "pairwise" or "poly". User-specified contrasts (or for that matter, any set of linear functions, be they contrasts or not) may be specified using a list of coefficients. Contrasts may also be requested directly from lsmeans via a contr argument or in the left-hand side of a formula, e.g., poly ~ dose | treat would request orthogonal polynomial contrasts of dose means at each level of each treat. The contrast function returns an lsmobj object; thus it is possible to do further analyses of those results, such as contrasts of contrasts.

Other useful methods for lsmobj objects include test and confint, which simply call summary with the implied portion of the statistical output; pairs, which calls contrasts for pairwise comparisons; cld, which provides a compact letter display of comparisons; glht and as.glht, which interface with the multcomp package (Hothorn, Bretz, and Westfall 2013) for more exacting multiplicity adjustments; and lsmip, which produces an interaction-plot-like display of the LS means.

lsmeans works as follows. First, if given a fitted-model object, the ref.grid is created. This entails reconstructing the dataset used in fitting the model, by calling a recover.data method. Then the factor levels and other summary information is used to define the reference grid, and an lsm.basis method is called to assemble other needed information, such as the linear function associated with each grid point, the regression coefficients, covariance matrix, degrees of freedom information, basis for estimable functions, and so forth. New recover.data and lsm.basis methods may be written to support additional model types. The ref.grid object contains all needed information needed for subsequent least-squaresmean analysis, independent of the model type. In mixed models fitted by a lme4 function, the

**pbkrtest** package (Halekoh and Højsgaard 2013), if installed, is used to adjust the covariance matrix and obtain degrees of freedom using the Kenward-Roger method. If degrees of freedom are not available, asymptotic results are used and labeled as such.

The lsmeans methods use the given specifications to obtain marginal averages of the linear predictors as needed, and the contrast function computes contrasts among the linear predictors. These altered sets of linear predictors define something quite similar, but more general, than a reference grid, outputted as an lsmobj object. The summary method does the statistical calculations; thus, one can re-summarize a result in a different way if needed.

There is also an lstrends function which uses a fitted model to obtain a difference quotient from two reference grids, and returns an lsmobj object. This is useful for comparing the slopes of lines in models where a covariate interacts with other predictors.

## 2. Some examples

Most of the remainder of this article consists of examples showing **lsmeans**'s features and how it can be used to advantage in a variety of situations.

## 2.1. Adjusted means in covariance models

Oehlert (2000), p.456, gives a dataset concerning repetitive-motion pain due to typing on three types of ergonomic keyboards. Twelve subjects having repetitive-motion disorders were randomized to the keyboard types, and reported the severity of their pain on a subjective scale of 0–100 after two weeks of using the keyboard. We also recorded the time spent typing, in hours. Here we enter the data, and obtain the plot shown in Figure 1.

It appears that hours and pain are linearly related (though it's hard to know for keyboard C), and that the trend line for keyboard A is higher than for the other two. To test this, consider a simple covariate model that fits parallel lines to the three panels:

```
R> typing.lm <- lm(pain ~ hours + keybd, data = typing)</pre>
```

The reference levels can be discerned by calling the ref.grid function:

```
R> ( typing.rg <- ref.grid(typing.lm) )
'ref.grid' object with variables:
   hours = 59
   keybd = A, B, C</pre>
```

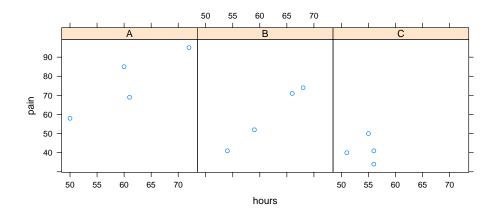


Figure 1: Display of the keyboard-pain data.

Note that only one variable has more than one level. Thus, the reference grid has only three points in it, corresponding to the three keyboards. The summary displays the predictions on this grid:

### R> summary(typing.rg)

```
hours keybd prediction SE df
59 A 73.565 3.6406 8
59 B 54.495 3.7223 8
59 C 49.440 3.9434 8
```

If we want lsmeans of the keyboard types, we get the same results, only by default, 95% confidence intervals are displayed:

```
R> ( typing.lsm <- lsmeans(typing.rg, "keybd") )</pre>
```

```
keybd 1smean
                  SE df lower.CL upper.CL
Α
      73.565 3.6406
                      8
                           65.170
                                    81.960
В
      54.495 3.7223
                      8
                           45.912
                                    63.079
С
      49.440 3.9434
                           40.346
                                    58.533
```

Confidence level used: 0.95

These results are the same as what are often called "adjusted means" in the analysis of covariance—predicted values for each keyboard, when the covariate is set to its overall average value.

The cov.reduce and at arguments can modify the reference grid. For example, by default, covariates are reduced to their means, but we can change this:

```
R> ref.grid(typing.lm, cov.reduce = median)
```

```
'ref.grid' object with variables:
  hours = 57.5
  keybd = A, B, C
```

Or we can use at to create a reference grid that contains more hours values:

```
R> typing.rg2 <- ref.grid(typing.lm, at = list(hours = c(50,60)))
R> lsmeans(typing.rg2, c("keybd", "hours"))
```

```
keybd hours 1smean
                        SE df lower.CL upper.CL
Α
         50 57.186 5.3185
                            8
                                44.922
                                          69.451
В
         50 38.116 5.5940
                                25.216
                                          51.016
                            8
С
         50 33.060 3.9434
                                23.967
                                          42.154
Α
         60 75.385 3.5944
                                67.096
                                          83.674
                            8
В
         60 56.315 3.6406
                            8
                                47.920
                                          64.710
C
         60 51.259 4.1093 8
                                41.783
                                          60.736
```

Confidence level used: 0.95

Again, these LS means are the same as the predictions at the six points of the reference grid. However, if we specify fewer predictors, we obtain marginal averages of the predictions:

R> lsmeans(typing.rg2, "keybd")

```
      keybd
      1smean
      SE
      df
      lower.CL
      upper.CL

      A
      66.286
      4.1548
      8
      56.705
      75.867

      B
      47.216
      4.3512
      8
      37.182
      57.250

      C
      42.160
      3.5886
      8
      33.885
      50.435
```

Results are averaged over the levels of: hours Confidence level used: 0.95

R> lsmeans(typing.rg2, "hours")

```
hours 1smean SE df lower.CL upper.CL 50 42.788 3.8865 8 33.825 51.750 60 60.987 2.1012 8 56.141 65.832
```

Results are averaged over the levels of: keybd Confidence level used: 0.95

Note that the results just above for keybd are not the same as the results we got the first time, using typing.rg. This illustrates the important point that *least-squares means depend* on the reference grid. In the first case, we have predictions at the average hours, 59, and in the second, we have the averages of predictions at 50 and 60 hours.

### 2.2. Follow-up analyses

There are several followup analyses available. Using our original typing.lsm result, we can obtain pairwise comparisons of them:

```
R> ( typing.pairs <- pairs(typing.lsm) )</pre>
```

```
      contrast
      estimate
      SE df t.ratio p.value

      A - B
      19.0699 5.0816 8 3.753 0.0138

      A - C
      24.1257 5.5596 8 4.339 0.0062

      B - C
      5.0558 5.7195 8 0.884 0.6647
```

P value adjustment: tukey method for a family of 3 means

Or the same results with a compact letter display (this requires that the **multcompView** package (Graves, Piepho, Selzer, and Dorai-Raj 2012) be installed):

```
R> cld(typing.lsm, alpha = .10)
```

```
keybd 1smean
                 SE df lower.CL upper.CL .group
C
      49.440 3.9434
                          40.346
                                   58.533
                      8
В
      54.495 3.7223
                     8
                          45.912
                                   63.079
                                           1
Α
      73.565 3.6406
                          65.170
                                   81.960
```

Confidence level used: 0.95

```
P value adjustment: tukey method for a family of 3 means significance level used: alpha = 0.1
```

In this display, two LS means that share at least one grouping symbol are not significantly different at the stated level. In this case, keyboard type A's predicted pain is significantly greater than either of the other two. By default, cld sorts the means, but this can be disabled.

Using the contrast function, other contrast families are available besides pairwise comparisons. For example, to obtain factor effects (differences from the grand mean), use:

```
R> contrast(typing.lsm, "eff")
```

```
contrast estimate SE df t.ratio p.value
A effect 14.3985 2.9954 8 4.807 0.0040
B effect -4.6714 3.0941 8 -1.510 0.1695
C effect -9.7271 3.3569 8 -2.898 0.0299
```

P value adjustment: fdr method for 3 tests

It is possible to provide custom contrasts as well—see the documentation.

Sometimes, we want to see different analyses of the same results. For example, the above results for pairs had a Tukey adjustment. If you want to know what the P values are with no adjustment, just do a different summary:

```
R> summary(typing.pairs, adjust = "none")
```

## 95% family-wise confidence level

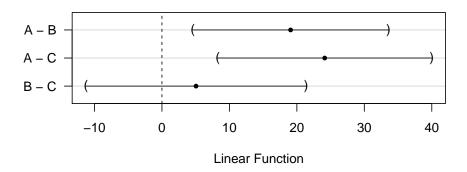


Figure 2: Graphical display of comparisons via multcomp

## 2.3. Interfacing with multcomp

As seen in the previous output, lsmeans provides for adjusting the p values of contrasts to preserve a familywise error rate. The default for pairwise comparisons is the Tukey (HSD) method. One must use these adjustments with caution. For example, when the standard errors are unequal, the Tukey method is only approximate, even under normality and independence assumptions. To get a more exact adjustment, we can convert an lsmobj object to a glht one for further analysis in the multcomp package (Hothorn et~al.~2013):

```
R> library("multcomp")
R> typing.glht <- as.glht(typing.pairs)
R> summary(typing.glht)
```

Simultaneous Tests for General Linear Hypotheses

#### Linear Hypotheses:

```
Estimate Std. Error t value Pr(>|t|)
A - B == 0
              19.07
                           5.08
                                   3.75
                                           0.0137 *
A - C == 0
              24.13
                           5.56
                                   4.34
                                           0.0062 **
B - C == 0
               5.06
                           5.72
                                   0.88
                                           0.6641
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
(Adjusted p values reported -- single-step method)
```

These p values are exact (if the assumptions hold) and, as expected, differ slightly from those in the previous 1smeans output. We may of course use other methods available for glht objects. The plot in Figure 2 displays the comparisons in the preceding table:

```
R> plot(typing.glht)
```

We have also provided an 1sm function that can be called within a glht call in a way similar to that of mcp as provided in the multcomp package. Here we display simultaneous confidence intervals for the LS means:

```
R> confint(glht(typing.lm, lsm("keybd")))
```

Simultaneous Confidence Intervals

```
Fit: lm(formula = pain ~ hours + keybd, data = typing)
```

```
Quantile = 2.953
```

95% family-wise confidence level

### Linear Hypotheses:

```
Estimate lwr upr
A == 0 73.565 62.815 84.316
B == 0 54.495 43.504 65.487
C == 0 49.440 37.795 61.084
```

The design of lsm is to create just one set of linear functions to hand to glht. It returns contrast output if specified, otherwise LS means output; so in the illustration above, the linear functions of the lsmeans themselves are used. If we had instead specified

```
R> lsm("keybd", contr="pairwise")
```

(output not shown) then the results would have been the same as shown earlier for the pairwise differences.

### 2.4. Fancy 1smeans calls

The lsmeans function allows for a lot of flexibility. we can call it with a fitted-model object instead of a ref.grid. If so, it can pass at and cov.reduce arguments to ref.grid. One may also specify contrasts and grouping variables. Here is an example:

```
R> lsmeans(typing.lm, specs = "keybd", by = "hours",
+ at = list(hours = c(50, 60)), contr = "trt.vs.ctrl1")
```

#### \$1smeans

```
hours = 50:
```

```
      keybd
      1smean
      SE df
      lower.CL
      upper.CL

      A
      57.186
      5.3185
      8
      44.922
      69.451

      B
      38.116
      5.5940
      8
      25.216
      51.016

      C
      33.060
      3.9434
      8
      23.967
      42.154
```

### hours = 60:

```
      keybd
      1smean
      SE df
      lower.CL upper.CL upper.CL

      A
      75.385
      3.5944
      8
      67.096
      83.674

      B
      56.315
      3.6406
      8
      47.920
      64.710

      C
      51.259
      4.1093
      8
      41.783
      60.736
```

Confidence level used: 0.95

 ${\tt P}$  value adjustment: sidak method for 2 tests

The result is a list with two lsmobj objects. When a by variable is present, the listings are grouped accordingly, and contrasts are restricted to each group.

In addition, a formula may be used in specs in place of all or part of the separate specs, by, and contr arguments. The following (not run) are all equivalent to the above:

```
R> lsmeans(typing.lm, specs = ~ keybd, by = "hours",

+ at = list(hours = c(50, 60)), contr = "trt.vs.ctrl1")

R> lsmeans(typing.lm, specs = ~ keybd | hours,

+ at = list(hours = c(50, 60)), contr = "trt.vs.ctrl1")

R> lsmeans(typing.lm, specs = trt.vs.ctrl1 ~ keybd | hours,

+ at = list(hours = c(50, 60)))
```

### 2.5. A three-factor experiment

The auto.noise dataset provided with Ismeans contains data from a factorial experiment wherein a newly design air-pollution filter called the Octel filter is compared with a standard filter with respect to the amount of ambient noise. Besides the factor type for which filter is used, the experiment includes three different sizes of cars (factor size) and measurements from each side of the car (factor side). First we fit a model to the data:

```
R> noise.lm <- lm(noise ~ size*type*side, data = auto.noise)
R> anova(noise.lm)
```

Analysis of Variance Table

Response: noise

```
Df Sum Sq Mean Sq F value Pr(>F)
                   26051
size
                2
                           13026 893.19 < 2e-16 ***
                    1056
                            1056
                                   72.43 1.0e-08 ***
type
                                   0.05 0.82910
side
                1
                       1
                               1
                2
                     804
                             402
                                   27.57 6.0e-07 ***
size:type
```

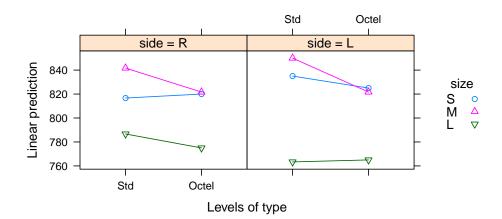


Figure 3: Three-way interaction plot for the auto.noise data.

```
44.33 8.7e-09 ***
size:side
                      1293
                               647
                                       1.19 0.28607
type:side
                 1
                        17
                                17
                                      10.33 0.00058 ***
size:type:side
                 2
                       301
                               151
Residuals
                24
                      350
                                15
Signif. codes:
                 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

The default reference grid for the 1smeans consists of all  $3 \times 2 \times 2 = 12$  factor combinations:

```
'ref.grid' object with variables:
    size = S, M, L
    type = Std, Octel
    side = R, L
```

R> ref.grid(noise.lm)

The model includes all interactions, so the LS means are the cell means. The **Ismeans** package provides a convenient function <code>lsmip</code> for displaying an interaction plot. (This feature requires **lattice** (Sarkar 2013) to be installed.) Figure 3 shows separate interaction plots for each side, via

```
R> lsmip(noise.lm, size ~ type | side)
```

The left side of the formula in lsmip specifies which factor(s) define the different curves, and the right side specifies the factor(s) for x axis. If a | character is included, it separates the plot into different panels. If two or more factors are given, their factor combinations are used to create a single factor for purposes of plotting. To illustrate, some variations on the plot in Figure 3 (not shown) are as follows:

```
R> lsmip(noise.lm, size ~ type * side) # 1 panel, 3 curves, 2*2 = 4 x values
R> lsmip(noise.lm, type * side ~ size) # 1 panel, 2*2 = 4 curves, 3 x values
R> lsmip(noise.lm, type ~ side | size) # 3 panels, 2 curves, 2 x values
```

The main goal of the experiment is to compare the mean noise levels for the two filters. One naïve way to do this is to simply ask for that comparison:

```
R> lsmeans(noise.lm, pairwise ~ type)
```

#### \$1smeans

```
type lsmean SE df lower.CL upper.CL Std 815.56 0.9001 24 813.70 817.41 Octel 804.72 0.9001 24 802.86 806.58
```

Results are averaged over the levels of: size, side Confidence level used: 0.95

#### \$contrasts

```
contrast estimate SE df t.ratio p.value
Std - Octel 10.833 1.2729 24 8.51 <.0001
```

Results are averaged over the levels of: size, side

```
Warning in lsmeans(noise.lm, pairwise ~ type) :
lsmeans of type may be misleading due to interaction with other predictor(s)
```

1smeans generates a warning message because the model includes interactions and it may not be wise to do main-effect comparisons. But whether it is wise or not, keep in mind that the LS means are marginal averages (using equal weights) of the predictions in the reference grid. So the LS mean for the Std filter is the average of the six predictions for which type = Std; and the LS mean for Octel is the average of the other six predictions. For a balanced experiment (which is the case here), these will be the same as the marginal means of the data:

```
R> with(auto.noise, tapply(noise, type, mean))
    Std Octel
815.56 804.72
```

So one way to look at marginal LS means for unbalanced data is that they are estimates of the marginal means we *would* obtain, had the experiment been balanced.

Now, given the strength of the interactions, it really is not smart to compare the marginal LS means for type; instead, we should compare them at each combination of the other factors. This is easily done by conditioning:

```
R> lsmeans(noise.lm, pairwise ~ type | size*side)[[2]]
size = S, side = R:
contrast estimate SE df t.ratio p.value
Std - Octel -3.3333 3.118 24 -1.069 0.2957
size = M, side = R:
```

```
SE df t.ratio p.value
contrast
             estimate
Std - Octel 20.0000 3.118 24
                                 6.414 <.0001
size = L, side = R:
contrast
             estimate
                         SE df t.ratio p.value
Std - Octel 11.6667 3.118 24
                                 3.742 0.0010
size = S, side = L:
             estimate
                         SE df t.ratio p.value
contrast
Std - Octel 10.0000 3.118 24
                                 3.207 0.0038
size = M, side = L:
contrast
             estimate
                         SE df t.ratio p.value
Std - Octel 28.3333 3.118 24
                                 9.087 <.0001
size = L, side = L:
contrast
             estimate
                         SE df t.ratio p.value
Std - Octel -1.6667 3.118 24
                               -0.535 0.5979
```

(We show only the second table of the results; the first table is the same as was shown earlier for the LS means of the three-factor combinations.) We find that in the four middle cases, the mean noise is statistically greater for the Std filter than the Octel filter. In the other two cases, the differences are nonsignificant. Note that a separate Tukey correction is made for each combination of the conditioning factors. Since each condition involves only two means, there is only one comparison and hence this amounts to no multiplicity correction at all. The conditioning also greatly reduces the output; if we had specified pairwise "type\*size\*side, we would have obtained estimates and tests of all  $\binom{12}{2} = 66$  pairwise comparisons of the 12 means, and the Tukey correction would have been based on 12 means also.

#### 2.6. Split-plot example

The nlme package includes a famous dataset Oats that was used in Yates (1935) as an example of a split-plot experiment. The dataset contains predictors Block (6-level factor), Variety (3-level factor), and nitro (4 unique numeric values). The experiment was conducted in six blocks, and each block was divided into three plots, which were randomly assigned to varieties of oats. With just Variety as a factor, it is a randomized complete-block experiment. However, each plot was subdivided into 4 subplots and the subplots were treated with different amounts of nitrogen. Thus, Block is a blocking factor, Variety is the whole-plot factor, and nitro is the split-plot factor. The response variable is yield, the yield of each subplot, in bushels per acre.

This experiment has random factors Block and Block: Variety (which identifies the plots). So we will fit a linear mixed-effects model that accounts for these. Another technicality is that nitro is a numeric variable, and initially we will model it as a factor. We will use lmer in the lme4 package (Bates et al. 2013) to fit a model:

```
R> data(Oats, package = "nlme")
R> library("lme4")
```

```
R> Oats.lmer <- lmer(yield ~ Variety*factor(nitro) + (1|Block/Variety),
+ data = Oats)
R> anova(Oats.lmer)
```

Analysis of Variance Table

```
      Variety
      2
      526
      263
      1.49

      factor(nitro)
      3
      20020
      6673
      37.69

      Variety:factor(nitro)
      6
      322
      54
      0.30
```

```
R> lsmip(Oats.lmer, Variety ~ nitro)
```

The interaction plot is displayed in Figure 4(a).

There is not much evidence of an interaction. Let's reduce to an additive model and look at the LS means and some appropriate contrasts

```
R> Oats.add <- lmer(yield ~ Variety + factor(nitro) + (1|Block/Variety),
+ data = Oats)
R> lsmeans(Oats.add, list(revpairwise ~ Variety, poly ~ nitro))
```

#### \$`lsmeans of Variety`

```
Variety 1smean SE df lower.CL upper.CL Golden Rain 104.500 7.7975 8.87 86.821 122.18 Marvellous 109.792 7.7975 8.87 92.113 127.47 Victory 97.625 7.7975 8.87 79.946 115.30
```

Results are averaged over the levels of: nitro Confidence level used: 0.95

#### \$`pairwise differences of Variety`

```
      contrast
      estimate
      SE df t.ratio p.value

      Marvellous - Golden Rain
      5.2917 7.0789 10 0.748 0.7419

      Victory - Golden Rain
      -6.8750 7.0789 10 -0.971 0.6104

      Victory - Marvellous
      -12.1667 7.0789 10 -1.719 0.2458
```

Results are averaged over the levels of: nitro P value adjustment: tukey method for a family of 3 means

#### \$`lsmeans of nitro`

```
nitro lsmean SE df lower.CL upper.CL 0.0 79.389 7.1324 6.64 62.335 96.442 0.2 98.889 7.1324 6.64 81.835 115.942 0.4 114.222 7.1324 6.64 97.169 131.276 0.6 123.389 7.1324 6.64 106.335 140.442
```

Results are averaged over the levels of: Variety

Confidence level used: 0.95

\$`polynomial contrasts of nitro`

```
contrast estimate SE df t.ratio p.value
linear 147.333 13.4395 51 10.963 <.0001
quadratic -10.333 6.0103 51 -1.719 0.0916
cubic -2.000 13.4395 51 -0.149 0.8823
```

Results are averaged over the levels of: Variety

The polynomial contrasts for nitro suggest that we could substitute a quadratic trend for nitro; so let's fit a third model where nitro is a quantitative predictor with a quadratic trend:

```
R> Oats.poly <- lmer(yield ~ Variety + poly(nitro, 2) + (1 | Block/Variety),
+ data=Oats)</pre>
```

If we want to see the same predictions as before, use the at argument to expand the reference grid:

```
R> Oats.poly.rg <- ref.grid(Oats.poly, at = list(nitro = c(0, .2, .4, .6)))
R> lsmeans(Oats.poly.rg, ~ Variety)
```

```
Variety lsmean SE df lower.CL upper.CL Golden Rain 104.500 7.7975 8.87 86.821 122.18 Marvellous 109.792 7.7975 8.87 92.113 127.47 Victory 97.625 7.7975 8.87 79.946 115.30
```

Results are averaged over the levels of: nitro Confidence level used: 0.95

R> lsmeans(Oats.poly.rg, ~ nitro)

```
    nitro
    lsmean
    SE
    df
    lower.CL
    upper.CL

    0.0
    79.289
    7.0923
    6.49
    62.249
    96.329

    0.2
    99.189
    6.8379
    5.62
    82.178
    116.199

    0.4
    113.922
    6.8379
    5.62
    96.912
    130.933

    0.6
    123.489
    7.0923
    6.49
    106.449
    140.529
```

Results are averaged over the levels of: Variety Confidence level used: 0.95

(Note: With the at argument omitted, we would obtain different LS means for Variety, because they would be predictions at the average nitro value of 0.3 rather than the averages of four predictions.) A simpler way to get the unique values of covariates is to specify cov.reduce = FALSE; we show this in a call to lsmip, which produces the interaction plot in Figure 4(b).

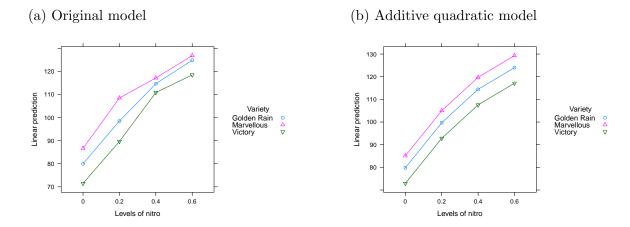


Figure 4: Interaction plots for the Oats experiment

R> lsmip(Oats.poly, Variety ~ nitro, cov.reduce = FALSE)

### 2.7. Messy data

To illustrate some more issues, and related lsmeans capabilities, consider the dataset named nutrition that is provided with the lsmeans package. These data come from Milliken and Johnson (1984), and contain the results of an observational study on nutrition education. Low-income mothers are classified by race, age category, and whether or not they received food stamps (the group factor); and the response variable is a gain score (post minus pre scores) after completing a nutrition training program.

Consider the model that includes all main effects and two-way interactions; and let us look at the group by race LS means:

```
R> nutr.lm <- lm(gain ~ (age + group + race)^2, data = nutrition)
R> lsmip(nutr.lm, race ~ age | group)
R> lsmeans(nutr.lm, ~ group*race)
```

group	race	lsmean	SE	df	lower.CL	upper.CL
${\tt FoodStamps}$	Black	4.7083	2.3681	92	0.0049714	9.4115
NoAid	Black	-2.1904	2.4906	92	-7.1368981	2.7561
${\tt FoodStamps}$	Hispanic	NA	NA	NA	NA	NA
NoAid	Hispanic	NA	NA	NA	NA	NA
${\tt FoodStamps}$	White	3.6077	1.1556	92	1.3125215	5.9028
NoAid	White	2.2563	2.3893	92	-2.4889667	7.0016

Results are averaged over the levels of: age Confidence level used: 0.95

Figure 5 shows the predictions from this model. One thing the lsmeans output illustrates is that lsmeans incorporates an estimability check, and returns a missing value when a prediction cannot be made uniquely. In this example, we have very few Hispanic mothers in the

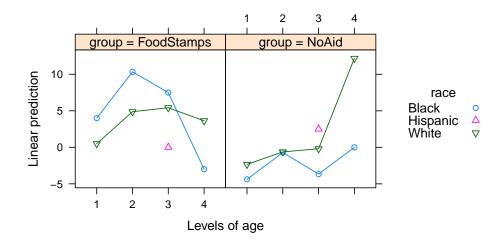


Figure 5: Predictions for the nutrition data

dataset, resulting in empty cells. This creates a rank deficiency in the fitted model and some predictors are thrown out.

We can avoid non-estimable cases by using at to restrict the reference levels to a smaller set:

```
R> lsmeans(nutr.lm, ~ group*race, at = list(age = "3"))
```

group	race	lsmean	SE	df	${\tt lower.CL}$	upper.CL
FoodStamps	Black	7.5000e+00	2.67205	92	2.1931	12.80693
NoAid	Black	-3.6667e+00	2.18172	92	-7.9998	0.66642
FoodStamps	Hispanic	2.1316e-14	5.34411	92	-10.6139	10.61386
NoAid	Hispanic	2.5000e+00	3.77885	92	-5.0051	10.00513
FoodStamps	White	5.4194e+00	0.95983	92	3.5130	7.32566
NoAid	White	-2.0000e-01	1.19498	92	-2.5733	2.17333

Confidence level used: 0.95

Nonetheless, the standard errors for the Hispanic mothers are enormous due to very small counts. One useful summary of the results is to narrow the scope of the reference levels to two races and the two middle age groups, where most of the data lie. However, always keep in mind that whenever we change the reference grid, we also change the definition of the LS means. Moreover, it may be more appropriate to average the two ages using weights proportional to their frequencies (23 and 64) in the data set. This may be done by changing the fac.reduce argument. With those ideas in mind, here are the LS means and comparisons within rows and columns:

So here are the results

#### R> nutr.lsm

```
lsmean
                               SE df lower.CL upper.CL
 group
            race
 FoodStamps Black 8.24896 2.9019 92
                                       2.4856 14.01234
            Black -2.88615 1.6914 92
                                      -6.2455
 FoodStamps White 5.27544 0.8649 92
                                       3.5577
                                               6.99322
 NoAid
            White -0.31236 1.0111 92 -2.3204
                                               1.69571
Results are averaged over the levels of: age
Confidence level used: 0.95
R> pairs(nutr.lsm, by = "race")
race = Black:
 contrast
                    estimate
                                 SE df t.ratio p.value
 FoodStamps - NoAid 11.1351 3.5444 92
                                         3.142 0.0023
race = White:
 contrast
                    estimate
                                 SE df t.ratio p.value
 FoodStamps - NoAid
                    5.5878 1.3305 92
                                         4.200 0.0001
Results are averaged over the levels of: age
R> pairs(nutr.lsm, by = "group")
group = FoodStamps:
 contrast
               estimate
                            SE df t.ratio p.value
                 2.9735 3.0047 92
 Black - White
                                    0.990 0.3250
group = NoAid:
               estimate
                            SE df t.ratio p.value
 contrast
 Black - White -2.5738 1.9706 92 -1.306 0.1948
```

Results are averaged over the levels of: age

The general conclusion from these analyses is that for age groups 2 and 3, the expected gains from the training are higher among families receiving food stamps. Note that this analysis is somewhat different than the results we would obtain by subsetting the data before analysis, as we are borrowing information from the other observations in estimating and testing these LS means.

#### 2.8. Trends

The **Ismeans** package provides a function **Istrends** for estimating and comparing the slopes of fitted lines (or curves). To illustrate, consider the built-in R dataset **ChickWeight** which has data on the growths of newly hatched chicks under four different diets. The following code produces the display in Figure 6.

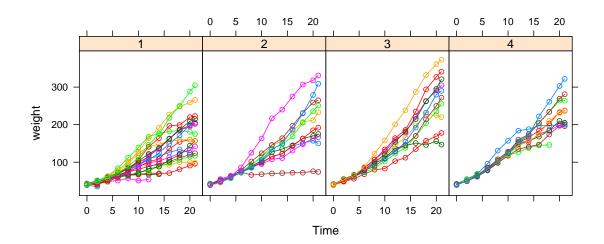


Figure 6: Growth curves of chicks, dataset ChickWeight.

Let us fit a model to these data using random slopes for each chick and allowing for a different average slope for each diet:

```
R> Chick.lmer <- lmer(weight ~ Diet * Time + (0 + Time | Chick),
+ data = ChickWeight)</pre>
```

We can then call lsmeans with a trend argument to estimate and compare the average slopes for each diet. Let's show comparisons of slopes using a compact letter display.

```
R> cld (lstrends (Chick.lmer, ~ Diet, var = "Time"))
```

```
Diet Time.trend
                      SE
                            df lower.CL upper.CL .group
1
         6.3386 0.61049 49.86
                                 5.1123
                                           7.5648
                                                  1
2
         8.6091 0.83800 48.28
                                 6.9245
                                          10.2938
                                                   12
4
         9.5558 0.83924 48.56
                                 7.8689
                                          11.2427
                                                    2
        11.4229 0.83800 48.28
                                 9.7382
                                         13.1075
```

Confidence level used: 0.95

P value adjustment: tukey method for a family of 4 means significance level used: alpha = 0.05

According to the Tukey HSD comparisons (with default significance level of .05), there are two groupings of slopes: Diet 1's mean slope is significantly less than 3 or 4's, Diet 2's slope is not distinguished from any other.

There is some additional trickery associated with trend. Consider the same model but with Time replaced by log(Time + 1):

```
R> Chick.lmer2 <- lmer(weight ~ Diet * log(Time + 1) +
      (0 + log(Time + 1) | Chick), data = ChickWeight)
R> cld (lstrends (Chick.lmer2, ~ Diet, var = "log(Time + 1)"))
Diet log(Time + 1).trend
                              SE
                                     df lower.CL upper.CL .group
                   43.101 3.8884 122.79
                                           35.404
                                                    50.798
                                                           1
2
                   58.493 5.3099 119.69
                                           47.979
                                                    69.006
                                                           12
 4
                   65.541 5.3366 121.74
                                           54.977
                                                    76.106
                                                             2
3
                   75.900 5.3099 119.69
                                           65.386
                                                    86.413
                                                             2
```

Confidence level used: 0.95

P value adjustment: tukey method for a family of 4 means significance level used: alpha = 0.05

This compares the trends that are fitted by the model. They compare in roughly the same way, but of course the values are much higher because the transformation has compressed the scale. But we can also look at the slopes for Time itself:

```
R> cld (lstrends (Chick.lmer2, ~ Diet, var = "Time"))
```

```
Diet Time.trend
                     SE
                             df lower.CL upper.CL .group
         3.6456 0.32889 122.79
                                  2.9946
                                            4.2967
                                                    1
2
         4.9475 0.44913 119.69
                                  4.0582
                                            5.8368 12
4
         5.5437 0.45138 121.74
                                  4.6501
                                            6.4373
                                                     2
3
         6.4198 0.44913 119.69
                                  5.5306
                                           7.3091
                                                     2
```

Confidence level used: 0.95

P value adjustment: tukey method for a family of 4 means significance level used: alpha = 0.05

These results are somewhat comparable to those we obtained with the first model. We will get a different set of slopes at different Times, because the fitted trends are curved with respect to Time.

### 2.9. Multivariate models

The MOats dataset provided in the package gives the Oats data mentioned previously, but with a multivariate response variable yield with four columns representing the yields of each plot with the four levels of nitrogen. We fit a model to these data

```
R> MOats.mlm <- lm(yield ~ Block + Variety, data = MOats)</pre>
```

This model assumes an unstructured covariance matrix on each plot. Here is its reference grid:

```
R> ref.grid(MOats.mlm)
```

```
'ref.grid' object with variables:
    Block = VI, V, III, IV, II, I
    Variety = Golden Rain, Marvellous, Victory
    rep.meas = multivariate response levels: 1, 2, 3, 4
The ref.grid function "flattens" the multivariate results by creating a pseudo-factor to ac-
count for the dimensions of the multivariate response. By default, the pseudo-factor is named
rep.meas with integer levels. It's often better to specify a more meaningful name and levels:
R > MOats.rg < ref.grid(MOats.mlm, mult.levs = list(nitro = c(0, .2, .4, .6)))
R> MOats.rg
'ref.grid' object with variables:
    Block = VI, V, III, IV, II, I
    Variety = Golden Rain, Marvellous, Victory
    nitro = multivariate response levels: 0.0, 0.2, 0.4, 0.6
Now we can obtain LS means and such just as we did previously
R> ( MOats.lsm <- lsmeans(MOats.rg, ~ nitro | Variety) )</pre>
Variety = Golden Rain:
 nitro lsmean
                   SE df lower.CL upper.CL
   0.0 80.000 5.5406 10 67.655
                                   92.345
   0.2 98.500 6.6020 10
                           83.790 113.210
   0.4 114.667 8.6954 10 95.292 134.041
   0.6 124.833 7.3032 10 108.561 141.106
Variety = Marvellous:
 nitro lsmean
                   SE df lower.CL upper.CL
   0.0 86.667 5.5406 10 74.321
                                   99.012
   0.2 108.500 6.6020 10
                           93.790 123.210
   0.4 117.167 8.6954 10
                          97.792 136.541
   0.6 126.833 7.3032 10 110.561 143.106
Variety = Victory:
 nitro lsmean
                   SE df lower.CL upper.CL
   0.0 71.500 5.5406 10 59.155
                                   83.845
   0.2 89.667 6.6020 10
                           74.956 104.377
   0.4 110.833 8.6954 10 91.459 130.208
   0.6 118.500 7.3032 10 102.227 134.773
Results are averaged over the levels of: Block
Confidence level used: 0.95
```

R> ( MOats.pcon <- contrast(MOats.lsm, "poly") )</pre>

## Variety = Golden Rain:

contrast estimate SE df t.ratio p.value linear 150.6667 24.2087 10 6.224 0.0001 quadratic -8.3333 9.5169 10 -0.876 0.4018 cubic -3.6667 31.9574 10 -0.115 0.9109

#### Variety = Marvellous:

### Variety = Victory:

contrast estimate SE df t.ratio p.value linear 162.1667 24.2087 10 6.699 0.0001 quadratic -10.5000 9.5169 10 -1.103 0.2957 cubic -16.5000 31.9574 10 -0.516 0.6169

Results are averaged over the levels of: Block

We can even obtain contrasts of contrasts to obtain interaction contrasts. In the following, we compare the polynomial contrasts among the varieties:

R> pairs(MOats.pcon, by = "contrast")

#### contrast = linear:

 contrast1
 estimate
 SE df t.ratio p.value

 Golden Rain - Marvellous
 21.5000 34.236 10 0.628 0.8085

 Golden Rain - Victory
 -11.5000 34.236 10 -0.336 0.9401

 Marvellous - Victory
 -33.0000 34.236 10 -0.964 0.6147

#### contrast = quadratic:

 contrast1
 estimate
 SE df t.ratio p.value

 Golden Rain - Marvellous
 3.8333 13.459 10 0.285 0.9565

 Golden Rain - Victory
 2.1667 13.459 10 0.161 0.9858

 Marvellous - Victory
 -1.6667 13.459 10 -0.124 0.9916

#### contrast = cubic:

 contrast1
 estimate
 SE df t.ratio p.value

 Golden Rain - Marvellous -17.8333 45.195 10
 -0.395 0.9184

 Golden Rain - Victory
 12.8333 45.195 10
 0.284 0.9567

 Marvellous - Victory
 30.6667 45.195 10
 0.679 0.7809

Results are averaged over the levels of: Block
P value adjustment: tukey method for a family of 3 means

### 2.10. GLMM example

The dataset cbpp in the lme4 package, originally from Lesnoff, Laval, Bonnet, Abdicho, Workalemahu, Kifle, Peyraud, Lancelot, and Thiaucourt (2004), provides data on the incidence of contagious bovine pleuropneumonia in 15 herds of zebu cattle in Ethiopia, collected over four time periods. These data are used as the primary example in lme4 for the glmer function, and it is found that a model that accounts for overdisperion is advantageous; hence the addition of the (1|obs) in the model fitted below. lsmeans may be used as in linear models to obtain marginal linear predictions for a generalized linear model or, in this case, a generalized linear mixed model.

```
R> cbpp$obs <- 1:nrow(cbpp)
R> cbpp.glmer <- glmer(cbind(incidence, size - incidence)
+ ~ period + (1 | herd) + (1 | obs), family = binomial, data = cbpp)</pre>
```

R> (cbpp.lsm <- lsmeans(cbpp.glmer, ~ period))</pre>

Here are the LSmeans for the four periods

```
period lsmean
                    SE df asymp.LCL asymp.UCL
                            -2.0819
1
       -1.5003 0.29671 NA
                                     -0.91868
2
       -2.7268 0.39924 NA
                            -3.5094 -1.94420
3
       -2.8291 0.41800 NA
                            -3.6485
                                     -2.00977
4
       -3.3665 0.53373 NA
                            -4.4128 -2.32031
```

Confidence level used: 0.95

These LSmeans are on the scale of the linear predictor, so the units are on the logit scale. If you want to see the predicted incidences, simply summarize the results and ask for "response" predictions:

R> summary(cbpp.lsm, type = "response")

```
        period
        prob
        SE df
        asymp.LCL
        asymp.UCL

        1
        0.182382
        0.044245
        NA
        0.110869
        0.285226

        2
        0.061411
        0.023012
        NA
        0.029047
        0.125187

        3
        0.055771
        0.022012
        NA
        0.025370
        0.118181

        4
        0.033358
        0.017210
        NA
        0.011976
        0.089455
```

Confidence level used: 0.95

The response predictions for certain contrasts come out on the odds-ratio scale:

#### 4 - 1 0.15470 0.091829 NA -3.1441 0.0050

P value adjustment: sidak method for 3 tests P values are asymptotic Tests are performed on the linear-predictor scale

When degrees of freedom are not available, as in this case, 1smeans emphasizes that fact by displaying NA for degrees of freedom and in the column headings.

## 3. Conclusions

**Ismeans** helps extend R's capabilities for the analysis of experimental data, especially for those users who have relied on SAS's least-squares means provisions. It goes beyond SAS in a few useful ways—for example, allowing for factor combinations even when an interaction is not in the model, and estimating trends. It provides a flexible and relatively simple way to obtain predictions from a linear model, or marginal averages thereof; and it also provides an extension of **multcomp**'s capabilities along these lines.

### References

- Bates D, Maechler M, Bolker B, Walker S (2013). *lme4*: Linear Mixed-effects Models Using Eigen and S4. R package version 1.1-0, URL http://lme4.r-forge.r-project.org/.
- Graves S, Piepho HP, Selzer L, Dorai-Raj S (2012). multcomp View: Visualizations of Paired Comparisons. R package version 0.1-5, URL http://CRAN.R-project.org/package=multcompView.
- Halekoh U, Højsgaard S (2013). **pbkrtest**: Parametric Bootstrap and Kenward Roger Based Methods for Mixed Model Comparison. R package version 0.3-8, URL http://CRAN.R-project.org/package=pbkrtest.
- Harvey W (1976). "Use of the HARVEY Procedure." In SUGI Proceedings. URL http://www.sascommunity.org/sugi/SUGI76/Sugi-76-20%20Harvey.pdf.
- Hothorn T, Bretz F, Westfall P (2013). *multcomp:* Simultaneous Inference in General Parametric Models. R package version 1.3-1, URL http://CRAN.R-project.org/package=multcomp.
- Lenth RV (2014). Ismeans: Least-Squares Means. R package version 2.00.
- Lesnoff M, Laval G, Bonnet P, Abdicho S, Workalemahu A, Kifle D, Peyraud A, Lancelot R, Thiaucourt F (2004). "Within-Herd Spread of Contagious Bovine Pleuropneumonia in Ethiopian Highlands." *Preventive Veterinary Medicine*, **64**, 27–40.
- Milliken GA, Johnson DE (1984). Analysis of Messy Data Volume I: Designed Experiments. Van Nostrand. ISBN 0-534-02713-7.
- Oehlert G (2000). A First Course in Design and Analysis of Experiments. W. H. Freeman.

Pinheiro J, Bates D, R-core (2013). nlme: Linear and Nonlinear Mixed Effects Models. R package version 3.1-113, URL http://CRAN.R-project.org/package=nlme.

Sarkar D (2013). *lattice*: Lattice Graphics. R package version 0.20-24, URL http://CRAN. R-project.org/package=lattice.

SAS Institute Inc (2012). "LSMEANS Statement." In SAS/STAT(R) 9.3 User's Guide. URL http://support.sas.com/documentation/cdl/en/statug/63962/HTML/default/ viewer.htm#statug\_introcom\_a0000003362.htm.

Yates F (1935). "Complex Experiments." Journal of the Royal Statistical Society (Supplement), 2, 181–247.

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